Chapter 19: Laboratory Support for the Surveillance of Vaccine-Preventable Diseases

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Surveillance for vaccine-preventable diseases requires the close collaboration of clinicians, public health professionals, and laboratorians. This chapter provides guidelines on which specimens to collect for each vaccine-preventable disease and how to interpret laboratory results.

Each professional dealing with vaccine-preventable diseases (VPDs) should identify sources of laboratory support for his or her clinical and public health practice. Table 1 lists all tests for VPDs and provides a convenient format for listing laboratories and laboratorians that can provide you with support. In addition to the guidelines presented in this chapter, state health department personnel (appendices 27-29) can provide additional guidance on specimen collection, transport, and other information.

I. General guidelines for specimen collection and laboratory testing

Specimen collection and shipping are the first steps in obtaining laboratory diagnosis or confirmation for VPDs. Many publications provide guidelines for specimen collection and laboratory testing for viral and microbiologic agents.[1-5] The CDC has compiled information on using the CDC laboratories as support for reference and disease surveillance (RDS).[6] This publication contains the form required for submitting specimens to the CDC (CDC 50.34) (Appendix 30) and information on general requirements for all specimens (Appendix 31). Although written to guide specimen submission to CDC, this publication as well as other guidelines for collecting, processing, storing, and shipping diagnostic specimens (Appendix 32) may be applicable to the submission of specimens to other laboratories.

II. Disease-specific guidelines for specimen collection and laboratory testing

This chapter summarizes information from Chapters 1-14 of this manual for quick reference. Table 2 lists confirmatory and other useful tests for the surveillance of vaccine-preventable diseases, and Table 3 summarizes specimen collection procedures for laboratory testing. Because some specimens require different handling procedures, be sure to check with the diagnostic laboratory prior to shipping. When in doubt about what to collect, when to collect, where to send specimens, or if you have other related questions, call the state health department and laboratory.

Diphtheria (see Chapter 1).

Isolates of *C. diphtheriae*, from any body site (respiratory or cutaneous), whether toxigenic or nontoxigenic, should be sent to the CDC Diphtheria Laboratory for reference testing at the direction of the state health department. To arrange specimen shipping, contact the state public health laboratory. A manual for the laboratory diagnosis of diphtheria has been published.[7]

Diagnostic tests used to confirm infection include **isolation** of *C. diphtheriae* and **toxigenicity testing**. Although no other tests for diagnosing diphtheria are commercially available, CDC and a few state public health laboratories can perform a **polymerase chain reaction (PCR)** procedure on clinical specimens to confirm infection even after treatment with antibiotics has begun.

A clinical specimen for isolation should be obtained from a case of respiratory diphtheria as soon as diphtheria is suspected, even if treatment with antibiotics has already begun.

- The laboratory should be consulted for appropriate transport of clinical specimens and alerted to the suspicion of diphtheria since isolation of *C. diphtheriae* requires special culture media containing tellurite.

- Specimens should be taken from the nose and throat and from the diphtheritic membrane.

- All suspected cases and their close contacts should have specimens taken from the nose and throat for culture. Culture of *C. diphtheriae* from close contacts may confirm the diagnosis of the case, even if the patient's culture is negative.

After *C. diphtheriae* has been identified in culture, the **biotype (substrain)** should be determined, using the isolate from culture.

Toxigenicity testing, using the **Elek test**, also should be performed to determine if the *C. diphtheriae* isolate produces toxin.

Measurement of the patient's serum **antibodies to diphtheria toxin** before administration of antitoxin may help in assessing the probability of the diagnosis of diphtheria.

- If antibody levels are low, diphtheria cannot be ruled out, but if levels are high, *C. diphtheriae* is unlikely to be the cause of illness.

PCR can detect non-viable *C. diphtheriae* organisms from specimens taken after antibiotic therapy has been initiated.

- Clinical samples for PCR (swabs, pieces of membrane, biopsy tissue) are best transported in silica gel sachets.

Haemophilus influenzae (see Chapter 2)

Diagnostic tests used to confirm infection include **isolation** of the organism from a normally sterile body site and detection of antigen in urine, blood, and cerebrospinal fluid (CSF).

Most hospital and commercial microbiologic laboratories have the ability to **culture** *H. influenzae* from clinical specimens. Normally sterile sites from which invasive *H. influenzae* may be isolated include CSF, blood, joint fluid, pleural fluid, pericardial fluid, peritoneal fluid, and subcutaneous tissue fluid.

Only disease due to *H. influenzae* **type b** (Hib) is vaccine preventable; thus, serotyping is of public health importance, as it indicates whether the case was vaccine-preventable, and may provide an opportunity to vaccinate susceptible contacts. Serotyping is not needed for clinical case management..

- To monitor the occurrence of invasive Hib disease, microbiology laboratories should perform serotype testing of all *H. influenzae* isolates from normally sterile sites, particularly those obtained from children <15 years of age.

Antigen detection (urine, blood, and CSF) may be used as an adjunct to culture, particularly in the diagnosis of patients who have received antimicrobial agents before laboratory specimens for culture were obtained.[8]

- If, in the absence of a positive sterile site culture, the Hib antigen is detected in the CSF, the patient should be considered a probable case of Hib disease and reported as such.

- Antigen detection tests can be positive in urine and serum of persons without invasive Hib disease, and persons who are identified exclusively by positive antigen tests in urine or serum should not be reported as cases.

Hepatitis A (see Chapter 3)

The diagnosis of acute hepatitis due to hepatitis A virus (HAV) is confirmed during the acute or early convalescent phase of infection by the presence of **IgM anti-HAV** in serum.

Serum for IgM anti-HAV testing should be drawn as soon as possible after onset of symptoms, since IgM anti-HAV generally disappears within 6 months after onset of symptoms.

During the convalescent phase of infection, **IgG anti-HAV** appears; it remains in serum for the lifetime of the person conferring enduring protection against disease.

The antibody test for **total anti-HAV** measures both IgG anti-HAV and IgM anti-HAV.

A test result of total anti-HAV positive and IgM anti-HAV negative indicates immunity consistent with either past infection or vaccination.

Hepatitis B (see Chapter 4)

Hepatitis B infection can be serologically confirmed either by a positive test for IgM antibody to hepatitis B core antigen **(IgM anti-HBc)** or by a positive test for hepatitis B surface antigen (HBsAg) with a negative test for anti-HAV (see Table 5).

IgM anti-HBc is present during acute HBV infection and may persist up to 6 months after onset of illness.

- Persons who test positive for total anti-HBc and negative for IgM anti-HBc have either resolved or chronic infection.
- A positive test for HBsAg with a negative test for IgM anti-HBc indicates chronic infection.
- Anti-HBc testing is not a reliable indicator of perinatal HBV infection because IgM anti-HBc is not detected in most infants with perinatal HBV infections and passively transferred maternal anti-HBc may persist beyond the age of 12 months.

Antibody to hepatitis B surface antigen (anti-HBs) indicates a response to either HBV infection or to hepatitis B vaccine. It can also be present among persons who have acquired anti-HBs passively (e.g., Hepatitis B immune globin administration).

- The primary use of anti-HBs testing is to determine if a vaccine recipient has responded to hepatitis B vaccine. For this purpose, testing should be performed between 1 and 2 months after completion of the vaccine series, except for infants born to HBsAg-positive mothers, in whom testing should be done between 9–15 months of age.

Hepatitis B surface antigen (HBsAg) for indicates active viral replication during acute and chronic infection.

Influenza (see Chapter 5)

Methods available for the diagnosis of influenza include **virus isolation** (standard methods and rapid culture assays), **detection of viral antigens** (enzyme immunoassays [EIA], immunoflourescent antibody [IFA], and less frequently electron microscopy), **molecular detection** (polymerase chain reaction [PCR]), and **serologic testing**.

When **direct antigen detection methods** are used to screen for influenza, it is important to save an aliquot of the clinical sample for further testing. These samples may be used for culture confirmation of direct test results and for subtyping influenza A isolates by the state public health laboratory. Full antigenic characterization of the viral isolate may be performed by the U.S. World Health Organization (WHO) Collaborating Center for Surveillance, Epidemiology and Control of Influenza, Influenza Branch, CDC. Full characterization is necessary for the detection and tracking of antigenic variants, an essential part of the selection of optimal influenza vaccine components.

Virus isolation is the gold standard for influenza diagnosis.

- Appropriate samples include nasal washes, nasopharyngeal aspirates, nasal and throat swabs, transtracheal aspirates, and bronchoalveolar lavage.

- To maximize the probability of isolating virus, samples should be taken within 72 hours of onset of illness.

- Rapid culture assays that detect viral antigens in cell culture are available. The results of these assays can be obtained in 18–40 hours as compared with an average of 4.5 days to obtain positive results from standard culture.

Viral antigen detection methods exist for the diagnosis of influenza infection directly from clinical material.

- Cells from the clinical sample can be stained using an immunofluorescent antibody to look for the presence of viral antigen. Nasal washes, nasopharyngeal aspirates, nasal and throat swabs, gargling fluid, transtracheal aspirates, and bronchoalveolar lavage are suitable clinical specimens.

- Commercially available kits to test for the presence of viral antigens fall into two groups; the first detects only influenza type A viruses, while the second detects both influenza type A and B viruses but does not differentiate between virus types. Results of these rapid antigen detection tests can be available in less than 1 hour.

- Other less frequently used methods include immunostaining and visualization of viral antigens by electron microscopy.

Molecular methods can be used to detect the presence of influenza virus in a clinical specimen and to characterize the virus. These methods include detection of viral RNA by molecular hybridization and reverse transcriptase PCR.

- when direct antigen detection methods are used for the diagnosis of influenza, it is important to collect and reserve an aliquot of the clinical sample for possible further testing. Reserved samples may be used for culture confirmation of direct test results and isolation for subtyping influenza A isolates.

- For some rapid testing methods, the media used to store the specimen is inappropriate for viral culture; in this case, it is necessary to collect two separate samples.

- Full antigenic characterization of the virus may be performed by the U.S. World Health Organization (WHO) Collaborating Center for Reference and Research on Influenza, Influenza Branch, CDC. Characterization of isolates is necessary for the detection and tracking of antigenic variants, an essential part of the selection of optimal influenza vaccine components. Molecular test procedures usually taken 2-3 days to complete.

Serologic diagnosis of influenza infection requires paired serum specimens. The acute sample should be collected within 1 week of the onset of illness and the convalescent sample should be collected approximately 2–3 weeks later.

- Hemagglutination inhibition tests are the preferred method of serodiagnosis. A positive result is a four-fold or greater rise in titer between the acute and convalescent samples when tested at the same time. Serologic test results are usually available in 24 hours.

- While serologic testing can be useful in certain situations where viral culture is not possible or in special studies, serologic diagnosis of influenza is not accepted for the purposes of national surveillance due to a lack of standardized testing methods and interpretation.

Measles (see Chapter 6)

Diagnostic tests used to confirm measles include serological tests for measles IgM antibodies and paired sera testing to assess the rise in IgG.[9,10] The IgM response will be transient (1–2 months) and the IgG response should persist. Thus, uninfected persons will be IgM negative and will either be IgG negative or IgG positive depending upon their previous infection histories (see Table 4).

Tests for **IgM antibody** require only a single serum specimen and are diagnostic if positive.

- There are two formats for IgM tests. The most widely available is the indirect format. IgM tests based on the indirect format require a specific step to remove IgG antibodies. Problems with removal of IgG antibodies can lead to false-positive tests, or less commonly, false-negative results.

- The second format, IgM capture, does not require the removal of IgG antibodies. These tests were developed at the CDC, and while these tests are not commonly available commercially, they are available at many state public health laboratories. This is the preferred reference test for measles and should be used to confirm every case of measles confirmed by any other laboratory test.

- If using the CDC IgM capture assay, tests which are negative in the first 72 hours after rash onset should be repeated on a second sera collected at least 72 hours after rash onset.

- Using the CDC IgM capture assay, IgM is detectable for at least 28 days after rash onset.

A variety of tests for **IgG antibodies to measles** are available.

- **IgG testing** for measles requires the demonstration of a four-fold rise in the titer of antibody against measles. Two serum specimens are always required. The first specimen should be drawn as soon after rash onset as possible, at the latest within 7 days after rash onset. The second specimen should be drawn 2-3 weeks later. The tests for IgG antibody should be conducted on both acute and convalescent specimens at the same time.

Although **isolation** of measles virus is not recommended as a diagnostic method, virus isolates are extremely important for molecular epidemiologic surveillance. Polymerase chain reaction (PCR) and other laboratory techniques are used to isolate the virus (Appendix 9).

- Specimens (urine, nasopharyngeal aspirates, heparinized blood, or throat swabs) for virus isolation should be obtained from every clinically suspected case of measles at the same time samples are taken for serologic testing.

- Because virus is more likely to be isolated when the specimens are collected within 3 days of rash onset, collection of specimens for virus isolation should not be delayed until laboratory confirmation is obtained.

- Clinical specimens should ideally be obtained within 7 days of rash onset, and should not be collected more than 10 days after rash onset.

Mumps (see Chapter 7)

Diagnostic tests that are used to confirm acute or recent mumps infection include **serologic tests** and **virus isolation.**

Mumps virus can be **isolated** from throat swabs, urine, and cerebrospinal fluid.

Mumps IgM antibodies are detectable within the first few days of illness, reach a maximum level about a week after onset of symptoms, and remain elevated for several weeks or months. False-positive IgM results by immunofluorescent antibody (IFA) assays have been reported.

Immunity to mumps may be documented by the presence of serum IgG antibodies. Since IgG antibodies may be detectable when the patient is first seen, a four-fold rise in IgG titer must be demonstrated for diagnosis. The acute serum specimen should be drawn within several days of illness onset. The convalescent specimen should be drawn at least 2 weeks later. The acute and convalescent serum specimens should be tested at the same time.

Pertussis (see Chapter 8)

Diagnostic tests that are used to confirm acute or recent pertussis infection include **isolation of *Bordetella pertussis*** and **polymerase chain reaction (PCR).**[11,12]

Isolation of *Bordetella pertussis* is the standard laboratory test for diagnosis of pertussis.

Whenever possible, suspected cases of pertussis should have a nasopharyngeal swab or aspirate obtained for **isolation of *B. pertussis*** . Among household contacts of culture-confirmed cases, diagnosis of pertussis is usually based upon a characteristic history and physical examination. Laboratory tests may be particularly useful for sporadic cases, for young infants, and in all cases modified by prior vaccination.

- Isolation of the organism using direct plating is most successful during the catarrhal stage (i.e., first 1–2 weeks of cough).

- Specimens from the posterior nasopharynx, not the throat, should be obtained using Dacron or calcium alginate (not cotton) swabs and should be plated directly onto selective culture media or placed in transport media (see figure in Chapter 8).

- Regan-Lowe agar or freshly prepared Bordet-Gengou medium is generally used for culture; half-strength Regan-Lowe can be used as the transport medium.

- Success in isolating the organism declines with prior antibiotic therapy effective against pertussis (erythromycin or trimethoprim-sulfamethoxazole), delay in specimen collection beyond the first 3 weeks of illness, or in vaccinated individuals.

- If antimicrobial testing or molecular typing (such as pulse-field gel electrophoresis) are indicated, bacterial isolates are required.

- Although isolation of *B. pertussis* is specific for the diagnosis, it is relatively insensitive. Under optimal conditions 80% of suspected cases in outbreak investigations can be confirmed by culture; in most clinical situations isolation rates are much lower.

DFA testing of nasopharyngeal secretions may be useful as a screening test for pertussis (a positive DFA result increases the probability that the patient truly has pertussis), but it is of limited specificity and should not be relied upon for confirmation.

Although **serological testing** has proven useful in clinical studies, it is not yet standardized. Due to lack of association between antibody levels and immunity to pertussis, results of serologic testing are difficult to interpret.

- In the absence of standardization, serologic test results should not be relied upon for case confirmation for the purposes of national reporting.

- Cases meeting the clinical case definition that are serologically positive, but **not** culture positive or PCR positive, should be reported as probable cases.

PCR testing of nasopharyngeal swabs or aspirates has been found to be a rapid, sensitive, and specific method for diagnosing pertussis. PCR, once validated in a laboratory, should be used in addition to culture, not as a replacement for culture, because bacterial isolates may be required for evaluation of antimicrobial resistance, or for molecular typing.

Pneumococcal infection (see Chapter 9)

Diagnosis of pneumococcal infection is confirmed by **culture** of *S. pneumoniae* from a normally sterile body site (e.g., blood, CSF, pleural fluid, or peritoneal fluid).

Based on recommendations from the National Committee for Clinical Laboratory Standards (NCCLS), all isolates of *S. pneumoniae* from normally sterile sites should be tested for penicillin resistance. Pneumococcal resistance to penicillin can be screened initially by using a 1 mg oxacillin disk; penicillin resistance is considered probable with oxacillin zone size <20 mm. The screening approach is highly sensitive (99%) and specific (80%–90%) and should detect nearly all isolates resistant to penicillin and extended-spectrum cephalosporins. Isolates found to be resistant by oxacillin disk should then be subjected to quantitative testing for other drugs that may be used to treat the patient.

Poliomyelitis (see Chapter 10)

Isolation of poliovirus (one of three serotypes) from the stool or pharynx early in the course of the disease is presumptive evidence of poliomyelitis. Isolation of virus from the cerebrospinal fluid (CSF) is diagnostic but is rarely accomplished.

The likelihood of poliovirus **isolation** is highest from stool specimens, intermediate from pharyngeal swabs, and very low from blood or spinal fluid.

- To increase the probability of poliovirus isolation, at least two stool specimens should be obtained 24 hours apart from patients with suspected poliomyelitis as early in the course of the disease as possible (i.e., immediately after poliomyelitis is considered as a possible differential diagnosis), but ideally within the first 15 days after onset of paralytic disease.

- Laboratories should forward isolates to CDC for intratypic differentiation to determine whether the poliovirus isolate is wild or vaccine-related.

Serology may be helpful in supporting or ruling out the diagnosis of poliomyelitis.

- An acute serum specimen should be obtained as early in the course of disease as feasible, and a convalescent specimen should be obtained 3 weeks later.

- A four-fold rise between the acute and convalescent specimens tested at the same time suggests poliovirus infection. Non-detectable antibody titers in both specimens may help rule-out poliomyelitis, but may be falsely negative in immunocompromised persons, who are also at highest risk for paralytic poliomyelitis.

- In addition, neutralizing antibodies appear early and may be at high levels by the time the patient is hospitalized; thus, a four-fold rise may not be demonstrated. One of the limitations of serology is the inability to distinguish between antibody induced by vaccine-related poliovirus and antibody induced by wild virus.

The **CSF** usually contains an increased number of leukocytes — from 10 to 200 cells/mm^3 (primarily lymphocytes) and a mildly elevated protein from 40 to 50 mg/100 ml. This finding is non-specific and may result from a variety of infectious and noninfectious conditions.

Rubella (see Chapter 11)

Diagnostic tests used to confirm acute or recent rubella infection or CRS include **serologic tests** or **virus isolation**.

Serologic tests used to confirm rubella include serological tests for rubella IgM antibodies and paired sera testing to assess the rise in IgG.[9,10] The IgM response will be transient (1–2 months) and the IgG response should persist. Thus, uninfected persons will be IgM negative and will either be IgG negative or IgG positive depending upon their previous infection histories.

- Sera should be collected as early as possible (within 7–10 days) after onset of illness, and again at least 7–14 days (preferably 2–3 weeks) later.

Rubella virus can be **isolated** from nasal, blood, throat, urine and cerebrospinal fluid specimens from rubella and CRS cases. Virus may be isolated from the pharynx 1 week before and until 2 weeks after rash onset.

Although **isolation** of the virus is diagnostic of rubella infection, viral cultures are labor intensive and are not done in many laboratories; they are not generally used for routine diagnosis of rubella. However, virus isolates are extremely important for molecular epidemiologic surveillance.[13] Specimens for virus isolation should be sent to CDC for molecular typing as directed by the state health department.

- Specimens (particularly pharyngeal swabs and urine specimens) for virus isolation should be obtained from every clinically suspected case of rubella at the same time as samples are taken for serologic testing.

- Because virus is more likely to be isolated when the specimens are collected within 4 days of rash onset, collection of specimens for virus isolation should not be delayed until laboratory confirmation is obtained.

- Clinical specimens should ideally be obtained within 4 days of rash onset (Appendix 17).

Congenital rubella syndrome (CRS) (see Chapter 12)

Laboratory confirmation can be obtained by any of the following:

- Demonstration of **rubella-specific IgM** antibodies in the infant's cord blood or infant's sera. In infants with CRS, IgM antibody persists for at least 6–12 months

- Documentation of the persistence of **serum rubella IgG titer** beyond the time expected from passive transfer of maternal IgG antibody

- **Isolation of the virus**, which may be shed from the throat and urine for a year or longer

- Detection of rubella virus by **polymerase chain reaction** (PCR)

Tetanus (see Chapter 13)

There are no laboratory findings characteristic of tetanus. The diagnosis is entirely clinical and does not depend upon bacteriologic confirmation. Sera collected before tetanus immune globulin is administered can demonstrate susceptibility of a patient to the disease.

Varicella (see Chapter 14)

Diagnostic tests used to confirm acute infection or immune status include **serologic tests**, **viral identification**, and **virus isolation**. Laboratory diagnosis of varicella is not routinely required, but may be useful in special circumstances such as cases of atypical clinical presentation or severe disease.

Serologic testing is available for detecting IgG and IgM antibodies to varicella zoster virus (VZV). Testing for IgM antibody is not indicated because available methods lack sensitivity and specificity. False positive IgM results are common in the presence of high IgG levels.

- Paired sera for IgG testing that show a four-fold rise in specific antibody titer may be used to confirm acute varicella infection.

- Acute sera should be collected within 7-10 days of rash onset; convalescent sera should be collected at least 7-14 days (preferably 2-3 weeks) later.

- Single IgG serology tests may be used to determine the immune status of a person who is a candidate for varicella zoster immune globulin (VZIG) or vaccination, but who has a negative or uncertain history of varicella.

A variety of methods have been used to detect IgG antibody to varicella zoster virus, both wild and vaccine strains. None of the tests listed below are as sensitive as the glycoprotein (gp) ELISA test used in pre-licensure clinical trials to study immune response post-vaccination however, this test is not commercially available. Currently available tests may not be sensitive enough to detect low levels of antibody post vaccination; however, the following tests may be useful:[14]

- Enzyme-linked immunosorbent assays (ELISA). The ELISA tests are widely used for screening of varicella immune status, especially when large numbers of specimens are tested. The ELISA tests range in sensitivity from 86% to 97% and range in specificity from 82% to 99% for detecting antibody after natural infection. Commercial ELISA are highly specific but less sensitive than the fluorescent antibody to membrane antigen test (FAMA) with the result that 10% to 15% individuals who are immune may be identified as susceptible.

- **Latex agglutination (LA)**. The LA test is useful for screening for varicella immunity. LA is rapid, simple-to-perform (15 minutes) assay to detect antibodies to VZV; dilutions of sera are added to latex particles coated with VZV glycoprotein antigen. It requires no specialized equipment, and has been reported to be nearly as sensitive as the FAMA, however, interpretation of borderline results is somewhat subjective. It is also less sensitive than FAMA in detecting antibody response following vaccination. No cross-reacting antibodies are detected by this technique, and false-positive reactions are rare.

- **Fluorescent antibody to membrane antigen (FAMA) test**. The FAMA test is highly sensitive, and is the "gold standard" for screening for immune status for VZV, but is not widely available. In convalescent-phase serum specimens, up to 100% are positive by FAMA and 96% by LA. After vaccination of persons who were previously VZV antibody negative, 77% are positive by FAMA, 61% by LA, and 47% by ELISA.

Rapid viral identification

Viral identification methods using direct fluorescent antibody techniques (DFA) may be used for rapid diagnosis of VZV infections; results may be obtained within several hours. Because viral proteins persist after cessation of viral replication, DFA may be positive when viral cultures are negative.

- Specimens for DFA testing should be collected by obtaining cells from the base of a lesion. After unroofing a fresh vesicle, swab the skin at the base of the lesion and rub onto a 6-8 mm area on a clean microscope slide. Allow to air dry.

- Other specimen sources that may be used include nasopharyngeal secretions, blood, urine, bronchial tree washing and cerebrospinal fluid;

however, these sources give lower positive yield compared with skin lesions.

Virus isolation

Virus isolation may be used to confirm diagnosis of VZV infections. Isolation of VZV, while difficult to perform, is diagnostic of varicella and zoster infection, and is indicated in cases of severe disease, especially in immunocompromised persons.

- Infectious VZV is most likely to be isolated from early vesicles with clear fluid rather than cloudy or crusted lesions.
- VZV is usually recoverable from varicella lesions for 2-3 days after first appearance of the lesions but can be isolated from zoster lesions for 7 days or longer.
- Other specimens for culture that give lower positive yield compared with skin lesions include nasopharyngeal secretions, blood, urine, bronchial tree washings and cerebrospinal fluid.
- Specimens for culture should be kept on dry ice or frozen at -70° C or below if storage for more than a few hours is required.

Virus typing or strain identification is labor-intensive and time-consuming. Few laboratories in the US have the technical capability for this testing.

- These tests should only be undertaken in special circumstances, most specifically to distinguish wild type from vaccine (Oka/Merck) virus.
- Merck and Co., Inc., offer a free viral identification service using polymerase chain reaction (PCR) and restriction fragment length polymorphism (RFLP) analysis and encourage its use in the following situations post vaccination: 1) rash with > 50 lesions; 2) suspected secondary transmission of the vaccine virus; 3) herpes zoster; or 4) any serious adverse event. Details of this service and the protocol for specimen collection can be obtained from Merck and Co., Inc. at 1-800-672-6372.

III. Public health action for positive laboratory reports

Many states and other jurisdictions have laws and/or regulations that require the reporting of certain laboratory results directly from the laboratory to the public health authorities.[15] Because physicians and other providers often do not report cases, the laboratory report may be the only report received by public health authorities.

Delays from symptom onset to initial case investigation often occur for cases which are reported to public health agencies through laboratory-based reporting. Because the laboratory rarely has the patient's address (or many other patient identifiers) and is not responsible for case investigations, delays in reporting may result. If reports are not received promptly, their usefulness as a trigger for public health action is diminished.

Even when public health action specifically related to a case may not be indicated (e.g., due to long lag time in reporting), situations where cases are not reported by clinicians but are later reported by a laboratory may serve to alert public health professionals to a weakness in their surveillance systems. Follow up with the health-care providers for these cases may enhance reporting by those clinicians. Close communication between the laboratorians, the clinicians, and the public health authorities can ensure that reports are received as needed for timely case investigation and public health intervention.

IV. Helpful phone numbers for more information

Additional information is available from the state public health department. For more information on use of laboratory testing in vaccine-preventable disease surveillance, contact your State Immunization Program (Appendix 27), State Epidemiologist (Appendix 28), or State Laboratory Director (Appendix 29). ❖

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